# Pea heat stress-responsive transcriptome analysis and heat tolerance improvement through marker-assisted backcrossing

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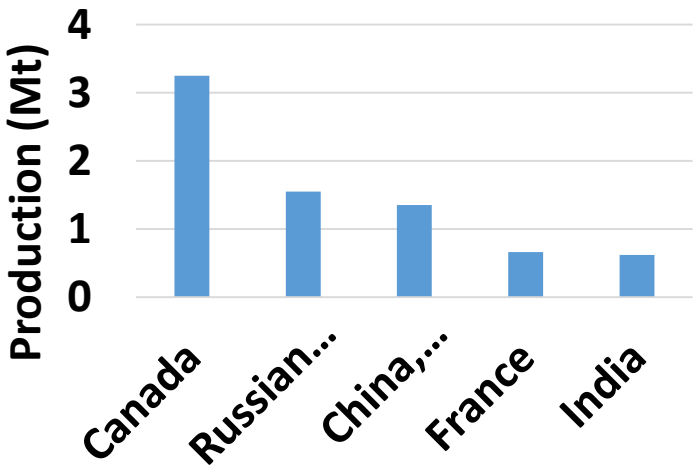
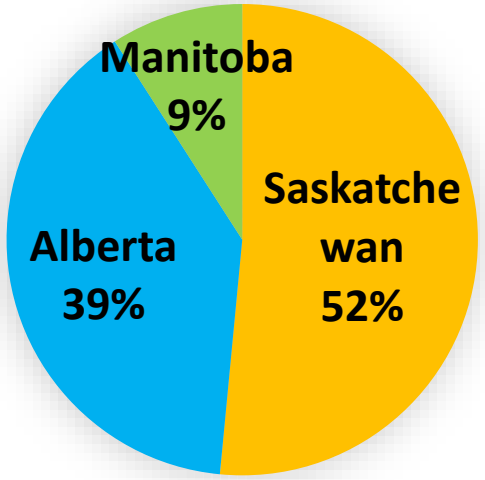
# Economic importance of dry pea in Canada

**Top five dry pea producing countries**




Data Source: FAOSTAT 2014 Mt: Million tons

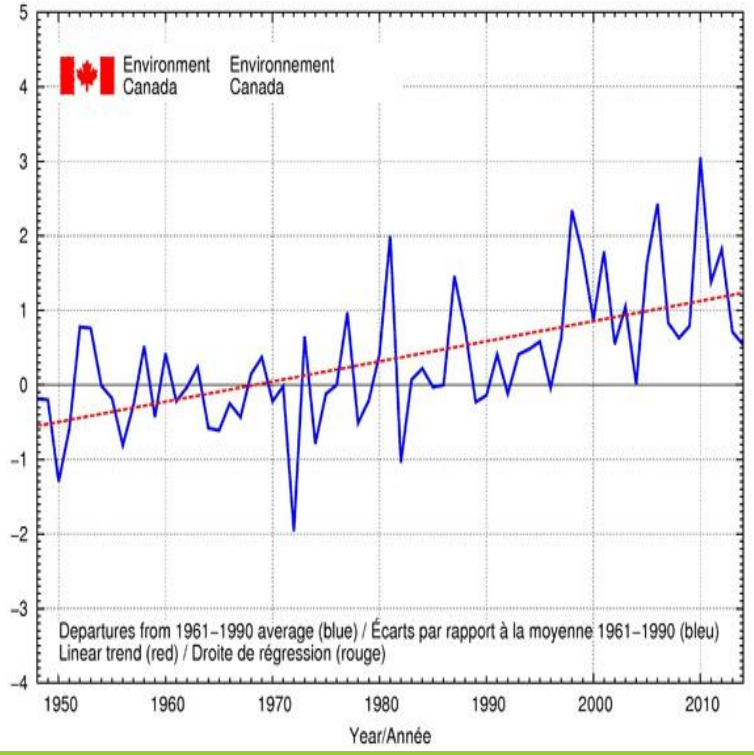
**Canada pea production distribution in 2016**



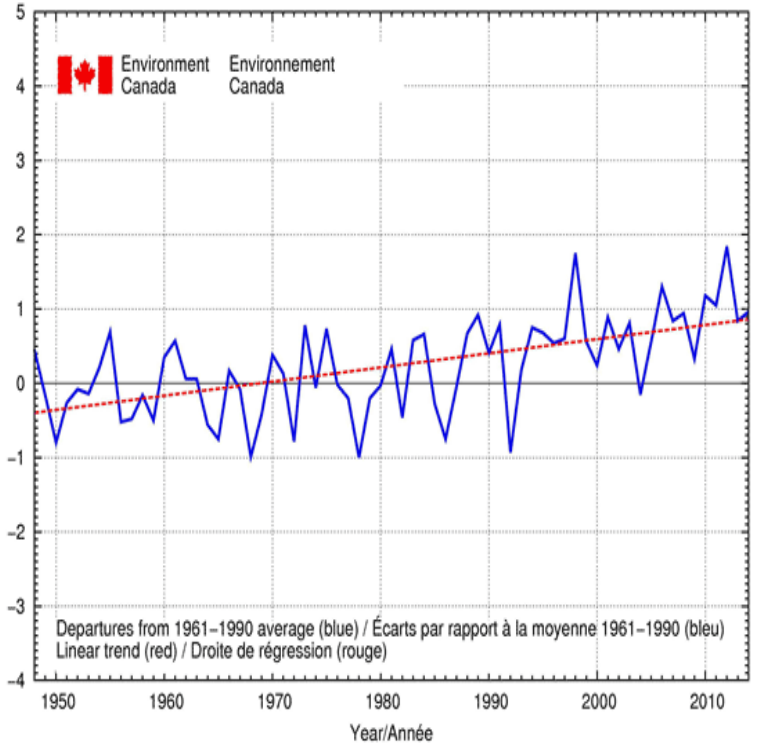

Data source: Agriculture and Agri-Food Canada

# Warming Canada

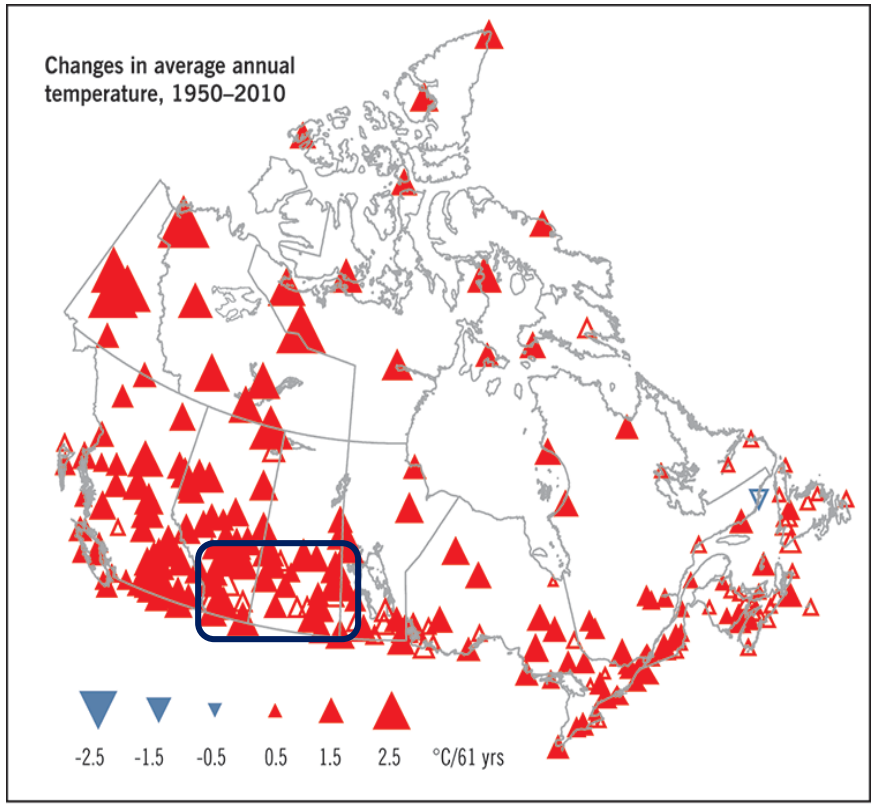
## Canada mean annual temperature




## Canada summer seasonal temperature



# Warming Canada

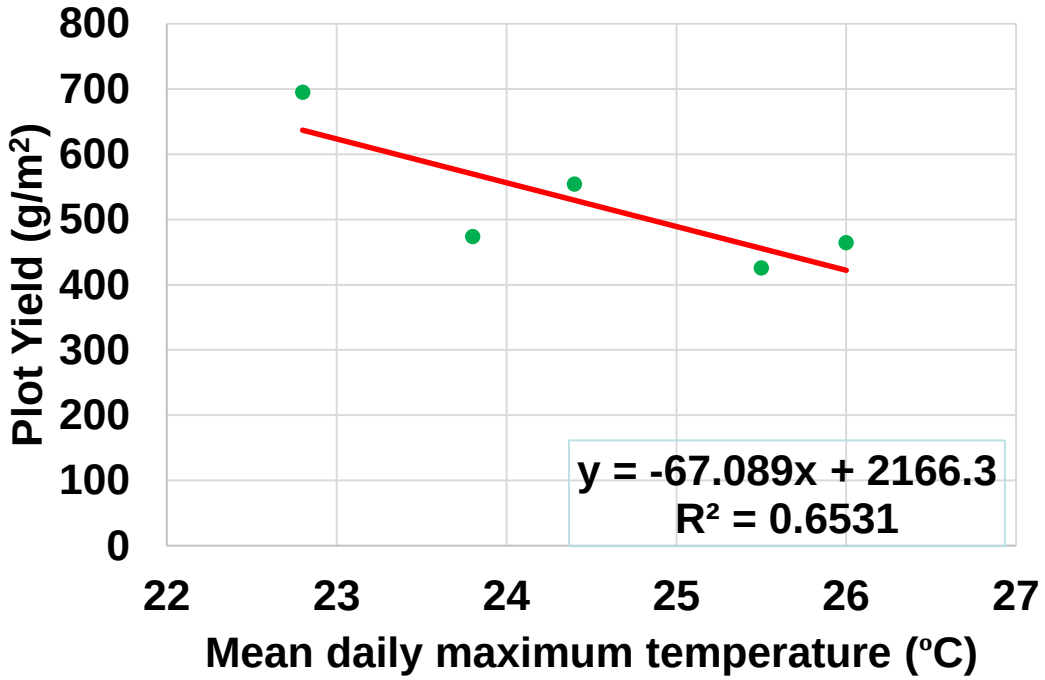




Source: Vincent et al (2012) Environment Canada

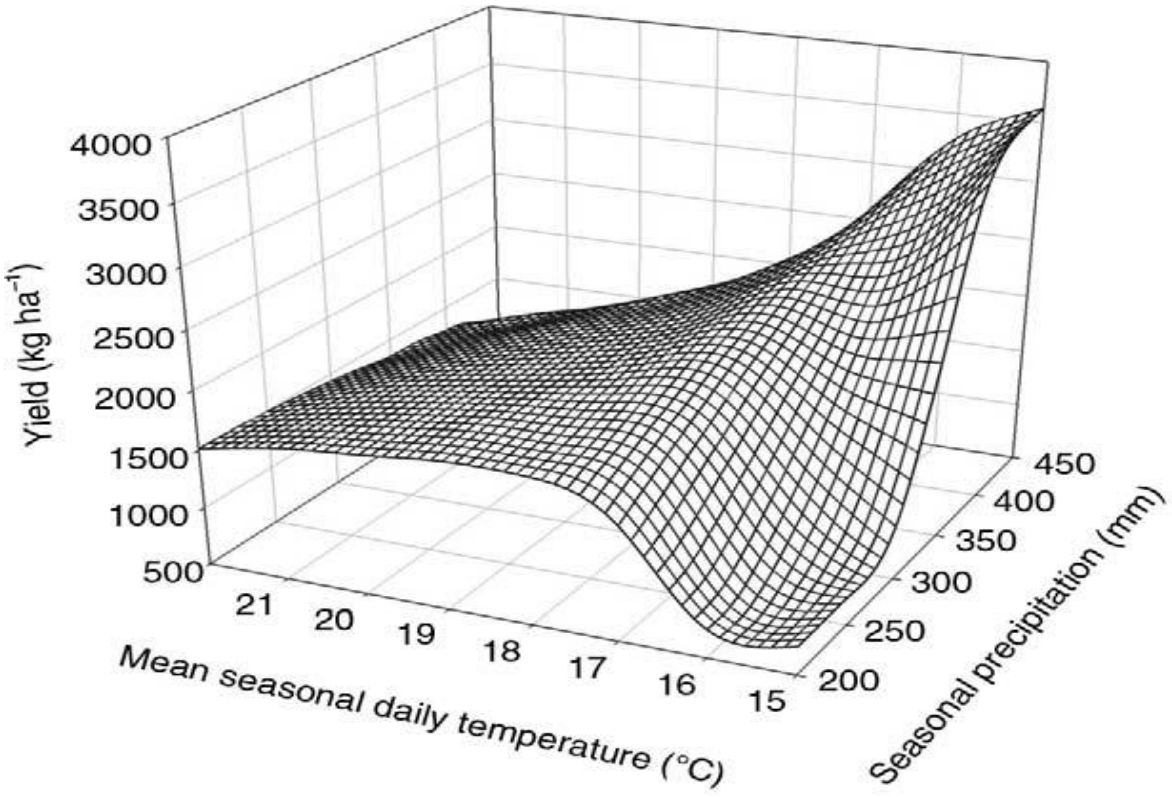
Saskatchewan Pulse Grower

# Pea Yield Response to Temperature




Huang, 2016

# Pea Yield Response to Temperature and Precipitation




Bueckert et al. 2015. Can. J. Plant. Sci

# Heat Stress on Pea

**Shortens life cycle** (Huang et al., 2017)

**Reduces pollen viability** (Jiang et al., 2015)

**Induces flower, ovule and pod abortion** (Jiang, 2016)

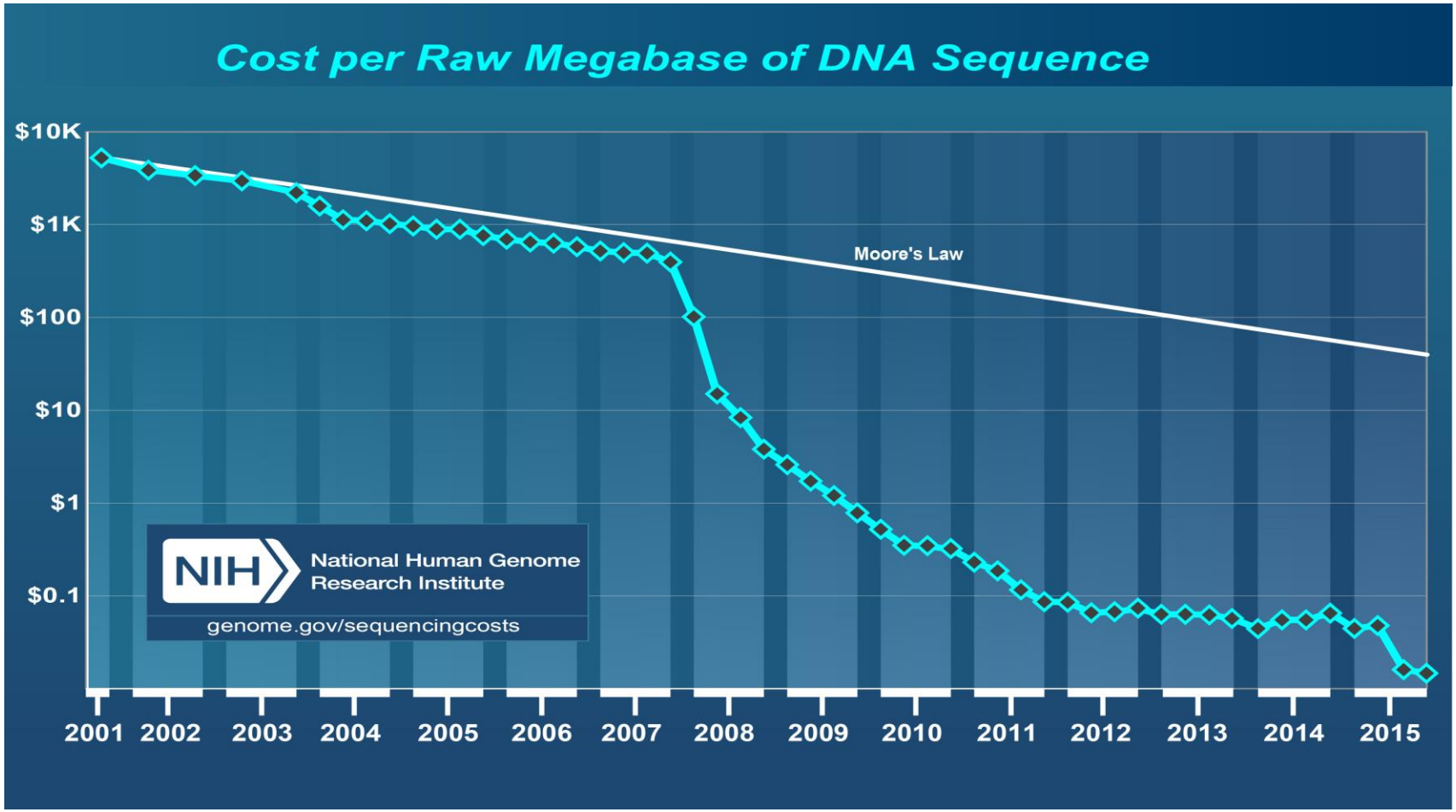
**Lowers individual seed weight** (Huang et al., 2017)

# Discovery of Heat Responsive Genes via Transcriptome Profiling

1. ***A. thaliana*** (Swindell et al., 2007)**; Wheat** (Qin et al., 2008); **Barley** (Mangelsen et al., 2011); **Canola** (Yu et al., 2014).

2. **Genes coding heat shock protein, heat shock factor, reactive oxygen species, primary and secondary metabolism.**

3. **The expression of heat responsive genes vary among different species and among different organs within a species.**


Cost per Raw Megabase of DNA Sequence
$10K
$1K
$100
$10
$1
$0.1
Moore's Law
NIH
National Human Genome Research Institute
genome.gov/sequencingcosts
2001
2002
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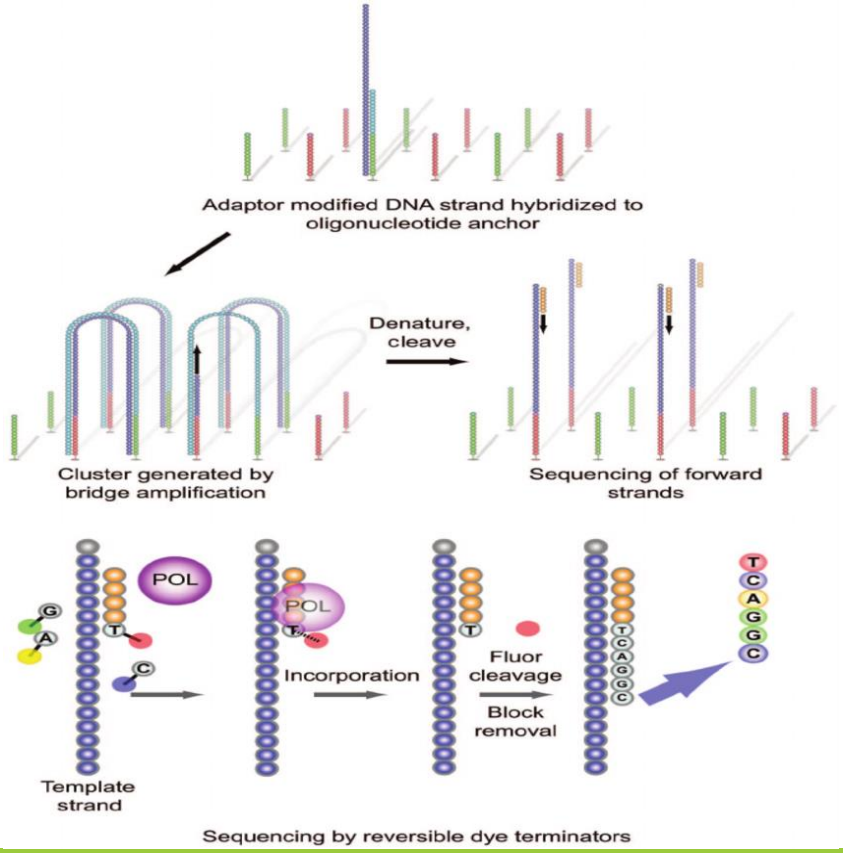
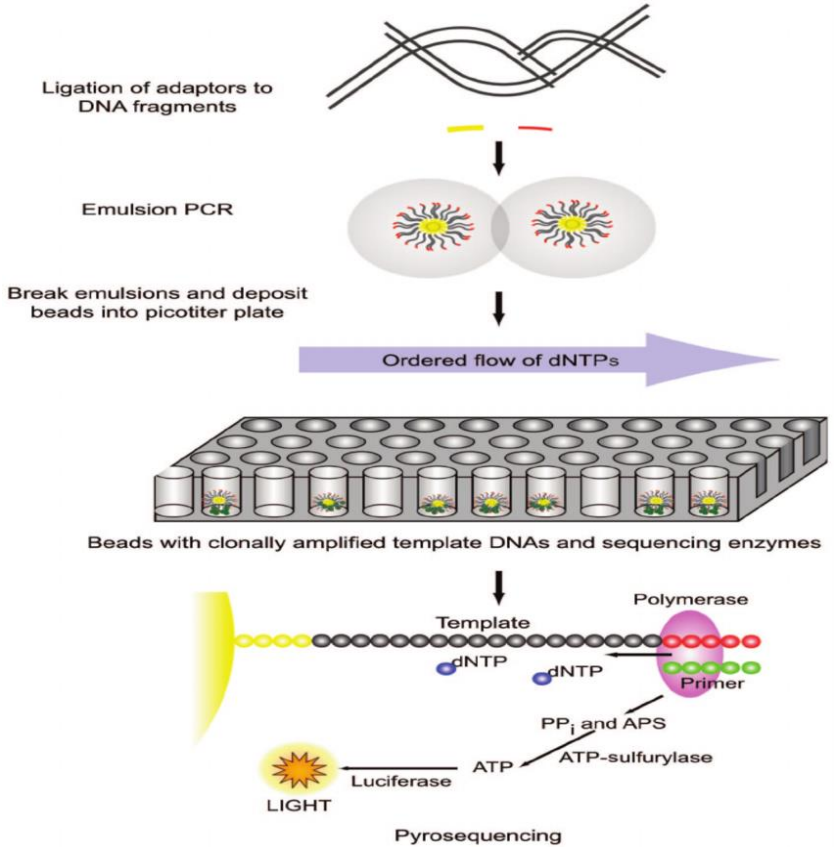
# Two Major Next-generation Sequencing Techniques

## Illumina




## Roche 454




Voelkerding et al., 2009

# Experiment 1: Identification of heat responsive genes in pea via transcriptional analysis

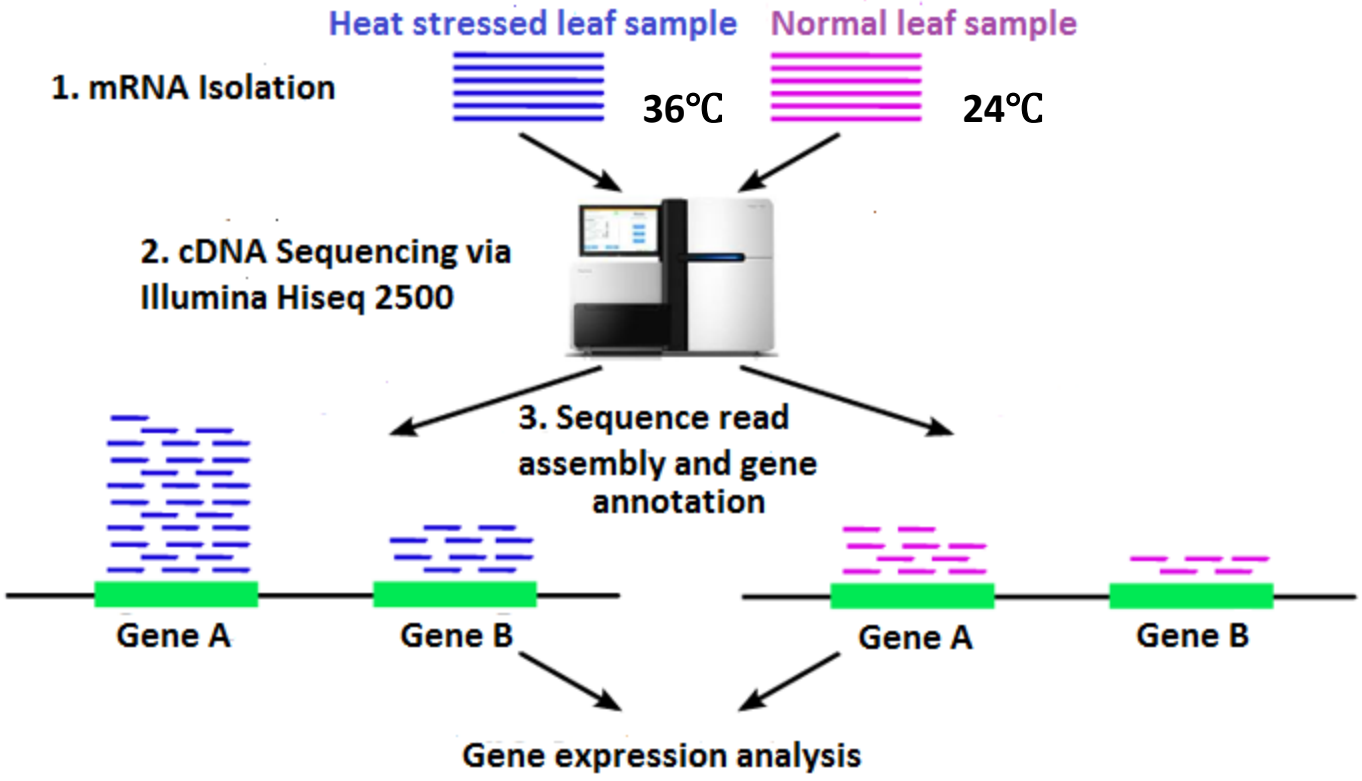
# Objectives:

1. Profile pea leaf response to high temperature at the transcription level via Illumina based RNA-seq

2. Compare the heat responsive gene expression between a heat tolerant (CDC Meadow; Warkentin et al., 2007) and a heat sensitive (Nitouche) pea variety

3. Build a heat responsive gene expression atlas in pea

# Experiment Protocol



# Experiment 2: Heat tolerance improvement via marker assisted backcrossing

# Plant Materials:

PR-11-2 ♂ X CDC Amarillo ♀

F1

⊗

F2 X CDC Amarillo ♀

MAS BC1 X CDC Amarillo ♀

MAS BC2 X CDC Amarillo ♀

MAS BC3 X CDC Amarillo ♀

BC4

⊗

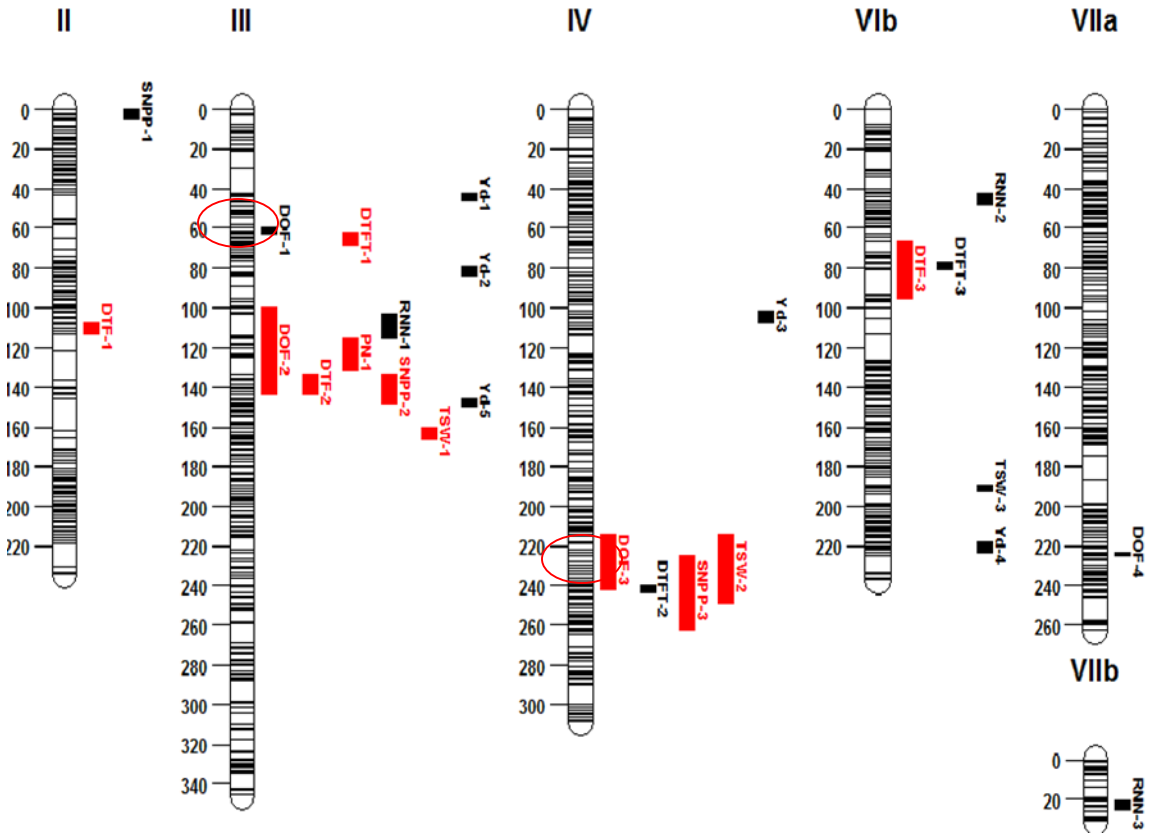
BC4F1

# Genome-wide SNP Markers

1. 1536 GoldenGate SNPs were identified (Sindhu et al., 2014).

2. Three hundred and fifty markers were selected based on the polymorphism between cultivated P. sativum including (CDC Sage and CDC Centennial – Parents of PR 11-2) map positions and converted to KASP assays and validated (Warkentin's lab, unpublished).

3. >50% of the KASP markers were polymorphic between PR11-2 and CDC Amarillo (Kishore et al., unpublished).




Huang et al. 2017

# Objectives:

1. Introgress loci conferring long flowering duration from PR-11-2 to CDC Amarillo

2. Identify QTL for heat tolerance between PR-11-2 and CDC Amarillo via the development of the backcrossing population

# Field Experiment:

**Randomized complete block design with 2 reps**

**Normal seeding date and Late seeding date**

**Traits of interest: days to flowering, flowering duration, reproductive node number, pod number, thousand seed weight, yield**

# Funding:


Western
Grains
Research Foundation


SASKATCHEWAN
pulse
Growers


Government
of
Saskatchewan